11. (Amended) The method of claim 1, where the modified living bacteria provided are additionally modified to be grown selectively.

12. (Amended) The method of claim 11, where the modified living bacteria grow selectively due to antibiotic resistance.

REMARKS

Claims 1-22 are pending in this application. Claims 13-22 were previously withdrawn from consideration. Claims 1-12 stand rejected under 35 U.S.C. §112. In response, claims 13-22 have been canceled, and claims 1-3 and 6-12 have been amended. No new matter is added by these amendments. Entry of these amendments is hereby requested.

With Respect to the Election/Restrictions, Paragraph 1 of the Outstanding Office Action:

Claims 1-22 were subjected to a Restriction Requirement dated March 28, 2002. An oral election of Group I, claims 1-12 was made by telephone conference with the Examiner on April 8, 2002. Claims 13-22 are canceled by this Response and Amendment.

With Respect to the Drawings, Paragraph 2 of the Outstanding Office Action:

The drawings stand objected to for the reasons set forth in the PTO 948 form attached to the Office Action. The form indicated that the figures considered were filed on January 25, 2003. However, formal figures were previously submitted on March 23, 2001, and were, apparently not forwarded to the draftsman. Copies of the formal figures previously submitted on March 23, 2001 are attached, as is a copy of the return post card from the United States Patent and Trademark Office acknowledging receipt of the formal figures March 26, 2001. Therefore, removal of this objection is hereby requested. If necessary, the Applicant will refile the formal figures.

With Respect to the Paragraph 4 of the Outstanding Office Action:

The Patent and Trademark Office has requested that trademarks given in the specification be capitalized and accompanied by the generic terminology, specifically identifying "MAXI-PREP KIT™ and BIORAD™" from page 8, lines 24. With respect to the Maxi-Prep DNA purification kit (Qiagen GmbH, Santa Clarita, CA, US) given on page 8, lines 16 and 24, the Applicant has been unable to locate any indication that the manufacturer Qaigen considers this name to be a trademark. Attached is a page from the manufacturer's website listing their marks. Additionally, the Applicant has performed an internet search and has been unable to locate any site discussing the kit that treats the name as a trademark or that uses the ™ next to the name. Therefore, the Applicant does not consider this name to be a trademark. With respect to generic terminology for a Maxi-Prep DNA purification kit, the kit contains syringes, reagents, buffers and several pieces of lab equipment. According to the undersigned's inquiry of the named inventor on this application, Shahrokh Shabahang, the name "Maxi-Prep DNA purification kit," or a similar identification, is the way this kit is referred to by those in this art and there is not another generic term useful for identifying the kit. If necessary, a declaration from Shahrokh Shabahang to this effect can be provided.

With respect to BIO-RAD® and BIO-RAD® GENE PULSER® II given on page 8, lines 18, 19 and 28, the disclosure has been amended to comply with the Patent and Trademark Office's requirement, and the identification of goods from Bio-Rad Laboratories, Inc.'s registered trademark for GENE PULSER®, United States Trademark Registration 1,485,127, has been added to the disclosure after the first occurrence of GENE PULSER®.

No other trademarks, registered or unregistered are apparent in the disclosure. Therefore, withdrawal of these objections is hereby requested.

With Respect to the rejections under 35 U.S.C. §112, First Paragraph, Paragraph 5 of the Outstanding Office Action:

All pending claims, claims 1-12, stand rejected under 35 U.S.C. §112, first paragraph, for the reasons discussed in paragraph 5 of the Office Action. Paragraph 5 contains

a series of rejections. The rejections will be addressed in the order they are given in the Office Action.

A) "The claims are so broad that they encompass evaluating every type of material, however, applicants have not described such a method. The instant specification fails to provide: the identity of a class of materials which can be evaluated; there is no discussion of whether the material can be solid or porous to allow the bacteria to pass through, around or into the material. If the material is antibiotic gauze or glass[,] how can one of skill in the art tell if the bacteria passed around or through the material?"

The Applicant respectfully traverses this rejection. The Patent and Trademark Office is correct; the claims do indeed encompass evaluating every type of material. That is the whole point of the invention; that any material, known now or in the future can be evaluated using this method. It would make no sense for a limitation to be placed in claim 1 requiring a specific type of material. If the properties of any particular material are known with respect to "whether [the] material will allow modified living bacteria to pass through the material or around the material or into the material," then using the method on a sample of the material is superfluous. Once a type of material is evaluated, presumably, the determination is made and that type of material would not need to be evaluated again. In any case, adding a limitation to claim 1 specifying one or more materials would render the invention moot and the Applicant declines to do so.

With respect to the two materials specified by the Patent and Trademark Office in the above passage, "antibiotic gauze" or "glass," one of ordinary skill in the art can tell "whether [the] material will allow modified living bacteria to pass through the material or around the material or into the material" by

"a) providing living bacteria which are modified to produce a first detectable signal;

b) placing the modified living bacteria on a first side of the [antibiotic gauze or glass] being evaluated; and

c) detecting whether the first signal is present on a second side of the [antibiotic gauze or glass] or within the [antibiotic gauze or glass];

where absence of the first signal on the second side of the [antibiotic gauze or glass] or within the [antibiotic gauze or glass] indicates that the modified living bacteria have not passed through or around the [antibiotic gauze or glass] and where presence of the first signal on the second side of the [antibiotic gauze or glass] or within the [antibiotic gauze or glass] indicates that the modified living bacteria have passed through or around the [antibiotic gauze or glass]," as claimed in claim 1, the same as any other material. If the material is very porous or not porous at all, it may be easy to predict that the modified bacteria will or will not pass into or through the material and that one will likely find the presence of the first signal on the second side of the material indicating that the modified living bacteria have passed through the material but it is irrelevant to the present method whether one or ordinary skill in the art can likely predict the determination being made by the method.

B) "If the bacteria dies [sic], as a result of being in contact with the material, one cannot say whether the bacteria passed through the material or not."

In response to this rejection, all instances of the word "bacteria" in the pending claim set have been amended to include the adjective "living." Support for this amendment can be found on page 5, lines 1-3, 6-7, 16-18, and page 6, line 29 through page 7, line 1, among other places.

C) "The method can only detect the modified bacteria; the method does not detect unmodified bacteria;..."

This assertion is correct. First, the method was invented specifically so it would **not** detect unmodified bacteria. As stated on page 1, lines 20-22, in the background section of the present application, there is a discussion of the prior art method of using bacteria (which prior art methods have not modified):

> "Though useful, this [prior art] method has several disadvantages . . . Finally, the presence of bacteria in the test medium can indicate that the testing apparatus itself was contaminated rather than that the material was breeched."

The present method detects only modified bacteria to distinguish modified bacteria that have "passed through the material or around the material or into the material" from unmodified bacteria that are present in the second side due to contamination of the material or testing apparatus.

Second, the first instance of the word "bacteria," after the transition phrase "comprising," in claim 1 as originally filed was in the limitation "providing bacteria which are **modified** to produce a first detectable signal" (emphasis added). All other pending claims, claims 2-12, depend on claim 1. Hence, it is clear that the claimed subject matter as originally filed refers only to "modified bacteria," not to unmodified bacteria. Further support for this limitation can be found in the disclosure at page 5, lines 1-12, among other places.

In response to this rejection, to remove any potential ambiguity, all other instances of the word "bacteria" in the pending claim set have been amended to include the adjective "modified." This amendment does not narrow the claimed subject matter as it was already present in the claims as originally filed.

D) "Moreover, if the modified bacteria fails[sic] to leave a fluorescent trail to demarcate its [sic] travel path, then one of skill in the art cannot assess where/how the bacteria traveled about the material; rather one can only say the bacteria did not attach itself [sic] to the material. Likewise, there is no teaching of how the bacteria will attach itself [sic] to the second side of the material in order to be detected. Does the material have some affinity for the bacteria or trap the bacteria based on size exclusion principles to determine whether the bacteria passed through, around or into the material? If the bacteria that fails [sic] to attach itself [sic] to the material but passes through, around or into the material, then the method steps are not commensurate in

scope to a method for evaluation of whether or not a material will allow bacteria to pass through, around or into the material."

First, the present method is not limited to modified bacteria that fluoresce. While fluorescence can be one signal that the bacteria can be modified to produce, and one example in the present application uses a type of fluorescence as a signal, the present method is not so limited. Fluorescence as a signal may be one embodiment of the present invention, even a preferred embodiment, but case law is well established that claims are not limited to a preferred embodiment or the best mode. Nor is the Applicant required to disclose a litany of potential signals, only to teach one of ordinary skill in the art how to make and use the invention in the best way known by the inventors as of the filing date. Clearly, anyone of ordinary skill in this art will understand that other signals, of types presently known, or of types to be invented in the future, can be used.

Second, "there is no teaching of how the bacteria will attach itself [sic] to the second side of the material" because attachment to the second side is not required. The present application does not contain the word "attach" even once. The idea that the present invention requires 'attachment of the bacteria to the second side of the material' is a complete mischaracterization of the invention.

Claim one as originally filed contains the following limitation (emphasis added):

> where absence of the first signal on the second side of the material or within the material indicates that the bacteria have not passed through or around the material and where presence of the first signal **on the second side of the material or within the material** indicates that the bacteria have passed through or around the material.

The word "side" in the phrase "on the second side of the material" is not synonymous with the word 'surface' on which something can be attached. As clearly disclosed and shown in the example on page 10, lines 13-15 and Figure 2, the signal generated by the modified bacteria is detected in the media on the second side of the material, not merely 'attached to the surface' of the material. A standard dictionary definition of "side" from Webster's Ninth New Collegiate Dictionary, a copy of which is attached, is as follows: side . .

. 1: the right or left part of the trunk of the body **2: a place, space, or direction with respect to a center or to a line of division** (as of an aisle, river, or street) 3 . . . b: a line or surface forming a border or face of an object . . . **4 . . . b: an area next to something . . .** If one says "I see my neighbor 'on the side' of his house, one does not mean that his neighbor's feet are attached to the side of his house and the neighbor's body is sticking out horizontally.

Therefore, there is no requirement that the present disclosure teach "how the bacteria will attach itself to the second side of the material."

E) "Moreover, the steps are so broad that it is unclear how to determine a first side or a second side of a material. For instance, if the material is root repair material and placed in the hollow center space of the tooth as discussed in the example at page 9, which side is the first and second side?"

A corresponding rejection was made in paragraph 6 of the Office Action and is addressed below.

F) "Also, the specification does not teach what amount of bacteria is required to be in contact with the material to determine whether the bacteria passed through, around or into the material."

This rejection is based on the same mischaracterization addressed in D) above. No amount of bacteria is required to be in contact with the material to determine whether the bacteria passed through, around or into the material.

G) "There is no description of the type of material that can or cannot be evaluated."

A corresponding rejection was made at the beginning of paragraph 5 of the Office Action and is addressed in A) above.

H) "It is unclear that one of skill in the art could use the claimed method to evaluate whether a material will allow bacteria to pass through the material or around the material."

The Applicant is mystified by this rejection. Obviously, one of skill in the art could use the claimed method because a detailed working example is provided, not merely a hypothetical example. All one of skill in the art would have to do is to read the disclosure and one would know that one could use the claimed method to evaluate whether a material will allow bacteria to pass through the material or around the material. If the Patent and Trademark Office persists in making this rejection, the Patent and Trademark Office is requested to state the reasoning for this rejection.

I) "The instant specification fails to provide a protocol that teaches method steps for such evaluation . . . "

The Applicant respectfully traverses this rejection. All that is necessary to teach one of ordinary skill in the art how to make and use the invention in the best way known by the inventors as of the filing date is provided in the disclosure, all of the essential method steps are recited in clam 1. The Patent and Trademark Office is requested to specifically identify what disclosure is insufficient or to withdraw this rejection.

J) "[F]urthermore, there is no data showing that such an evaluation will result in the determination of whether a material will allow a [sic] bacteria to pass through the material or around the material or into that material."

The Applicant respectfully traverses this rejection. There is no legal requirement that a disclosure in a patent application provide data to back up its application for a patent, only that the disclosure teach one of ordinary skill in the art how to make and use the invention in the best way known by the inventors as of the filing date. As discussed above, this legal requirement has been met. There is no additional disclosure needed.

The Patent and Trademark Office is requested to specifically identify what data is required to meet the legal requirement or to withdraw this rejection.

Therefore, all of these rejections are believed to be addressed by the claim amendments, or to be moot in view of the arguments made in this Response and Amendment. Withdrawal of all of these rejections is hereby requested.

With Respect to the rejections under 35 U.S.C. §112, second paragraph, Paragraph 6 of the Outstanding Office Action:

All pending claims, claims 1-12 stand rejected under 35 U.S.C. §112, second paragraph, for the reasons discussed in paragraph 6 of the Office Action:

> The terms "first side" [and] "second side" in the claims is [sic] relative terms, which render the claims indefinite. The terms are not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised [sic] of the scope of the invention. The metes and bound [sic] of the terms are broad thereby making the claims indefinite.

The Applicant respectfully traverses this rejection. As originally filed, claim one includes the recitations:

> "b) placing the bacteria on a first side of the material being evaluated; and
>
> c) determining whether the first signal is present on a second side of the material or within the material";

As stated, the "first side" is the side of the material where the modified living bacteria are placed. The "second side" is the side of the material where a determination is made whether the first signal is present. Support for this can also be found in the disclosure at page 5, lines 25-29, among other places.

The "side" of the material does not refer merely to a "surface" of the material, but a space bounded by a surface of the material that acts as a potential barrier between the first side and the second side. If the Applicant had wanted to limit its invention to a method for evaluating whether a material will allow bacteria to pass from 'one surface of a material to

another surface of the material,' it would have disclosed the invention in such a manner. However, the Applicant explicitly disclosed the invention in terms of a method for evaluating whether a material will allow bacteria to pass through the material or around the material from a first side to a second side or into the material. See for example, the specification at page 9, line 27 to page 10, line 18. The modified bacteria in liquid broth media was placed in a space in the tooth 14, the first side, not merely coated onto the surface of the tooth. The modified bacteria were detected in the media in the lower chamber 20, the second side, not on a surface of the tooth 14. Claims 9 and 10 as originally filed specifically claim methods based on this example. These claims could not be understood if the "first side" and the "second side" merely referred to a surface rather than a space. Thus, the"first side" and the "second side" are determined in the context of the method and are not limited to merely a surface of the material. Therefore, withdrawal of these rejections are hereby requested.

With Respect to the rejections under 35 U.S.C. §112, second paragraph, Paragraph 7 of the Outstanding Office Action:

All pending claims, claims 1-12 stand rejected under 35 U.S.C. §112, second paragraph, for omitting an essential step, for the reasons given in paragraph 7 of the Office Action:

> ...The omitted steps are: there is no detection step. The claims fail to positively recite how the first or second signal is detected on the second side of the material within the material. It is noted that determination of whether the first or second signal on the material is not equivalent to an actual detection step.

The Applicant respectfully traverses this rejection. As originally filed, and as noted by the Patent and Trademark Office, claim 1 includes the recitations:

> "c) determining whether the first signal is present on a second side of the material or within the material";

The Applicant believes that this recitation sufficiently contains the essential step identified by the Patent and Trademark Office. "Determining whether the first signal is present on a second

side of the material or within the material" includes detecting whether the first signal is present on a second side of the material or within the material. Notwithstanding the Applicant's belief, claims 1, 3, 9 and 10 have been amended to replace the word "determining" with the word "detecting." However, the Applicant states for the record that this amendment broadens the claim rather than narrowing the claim, as detecting is more specific than determining.

With respect to reciting "how the first or second signal is detected on the second side of the material within the material," the Applicant believes that there is no requirement that a specific detection method be recited, thereby limiting the method to any particular embodiment. As will be understood by those with skill in the art with reference to the present application, one of ordinary skill in the art could modify bacteria to have any of a number of detectable first signals and second signals requiring corresponding detection procedures. Thus, the present invention is not limited to merely one type of detection but the type of detection would vary with the type of the first signal and the second signal. Claims 2-12 depend on claim 1. Therefore, withdrawal of these rejections are hereby requested.

With Respect to the rejections under Paragraph 8 of the Outstanding Office Action:

Claim 1 stands rejected (presumably under 35 U.S.C. §112, second paragraph) for the reasons given in paragraph 8 of the Office Action.

> The preamble of claim 1 is drawn to a method for evaluating whether a material will allow a bacteria to pass through the material or around the material or into the material compromising [sic]:...however there [sic] the preamble of the claim must be drawn to the same method steps which are recited by the body of the claims. The claims are inconsistent since the body of the claim does not teach how to evaluate whether a material will allow bacteria to pass through, around or into the material, but rather determine whether the bacteria has [sic] attached itself to the material . . .

As discussed in the response to the rejections under 35 U.S.C. §112, first paragraph, paragraph 5, section D) above, claim one is not limited to determining "whether the bacteria has attached itself [sic] to the material" This is a mischaracterization of the invention. There is no support in the application for the Patent and Trademark Office characterization of claim 1

as requiring that the modified bacteria attach themselves to the material to be detected. Therefore, withdrawal of this rejection is hereby requested.

CONCLUSION

The Applicants believe that all pending claims, claims 1-12, are now in condition for allowance and an indication of such is requested. If, however, there remain any issues which can be addressed by telephone, the Examiner is encouraged to contact the undersigned.

The Commissioner is hereby authorized to charge payment of any fees associated with this communication to Deposit Account No. 19-2090.

Respectfully submitted,

SHELDON & MAK
a Professional Corporation

Date: April 14, 2003

By _____
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VERSION WITH MARKINGS TO SHOW CHANGES MADE

IN THE DISCLOSURE:

Page 8, lines 8-28:

Examples of the present method will now be described in greater detail. Modified bacteria containing genes to produce a functional green fluorescent protein, a functional luciferase and to contain an antibiotic resistance gene were constructed for use in the method by transformation with a plasmid DNA bearing a cassette with genes producing luciferase, green fluorescent protein and antibiotic resistance as follows. Two constructs were used. The first construct pLITE201 (as disclosed in Voisey CR, Marincs F. Biotechniques 1998;24:56) was a plasmid vector with a gram-negative origin of replication containing the *lux CDABE* cassette from *Xenorhabdus luminescens* driven by the lac promoter. It was purified from DH5α using the Maxi-Prep DNA purification kit (Qiagen GmbH, Santa Clarita, CA, US). The pLITE201 plasmid was then electroporated into attenuated strains of *Vibrio cholera*, *Salmonella typhimurium*, and *Shigella* using [BioRad®] BIO-RAD® electroporation protocols for the various strains and the [BioRad® Gene Pulser II] BIO-RAD® GENE PULSER® II unit (a pulse generator for transfectation of nucleic acids into mammalian, plant, or bacterial cells, Bio-Rad laboratories, Hercules, CA). Positive transformants were identified by placing the outgrowth plates under the Argus 100 low light imager (Hamamatsu Corp., Hamamatsu, Japan). The positive colonies were confirmed by observing fluorescent bacteria under the fluorescent microscope.

The second construct was a *lux ABCDE* cassette from pXylA-dual (Hill, P, University of Nottingham, UK) as shown in Figure 1, purified using the Maxi-Prep kit (Qiagen). This plasmid has a gram-positive origin of replication as well as gram-positive ribosomal binding sites, which allowed expression in gram-positive organisms. The plasmid was then transformed into *Enterococcus faecalis* (strains JH2-2, ATCC4082 , and OG1X) using

electroporation with the [BioRad® Gene Pulser II] BIO-RAD® GENE PULSER® II (Bio-Rad Laboratories, Hercules, CA).

IN THE CLAIMS:

Cancel claims 13-22:

Amend claims 1 as follows:

1. (Amended) A method for evaluating whether a material will allow modified living bacteria to pass through the material or around the material or into the material comprising:

a) providing living bacteria which are modified to produce a first detectable signal;

b) placing the modified living bacteria on a first side of the material being evaluated; and

c) [determining] detecting whether the first signal is present on a second side of the material or within the material;

where absence of the first signal on the second side of the material or within the material indicates that the modified living bacteria have not passed through or around the material and where presence of the first signal on the second side of the material or within the material indicates that the modified living bacteria have passed through or around the material.

2. (Amended) The method of claim 1, additionally comprising quantifying the amount of modified living bacteria that will pass through the material or into the material by quantifying the amount of the first signal on the second side of the material;

where increasing amounts of the first signal on the second side of the material or within the material indicates increasing amounts of modified living bacteria that will pass through the material or into the material.

3. (Amended) The method of claim 1, where the modified living bacteria are modified to produce a second detectable signal, and where the method additionally comprises [determining] detecting whether the second signal is present on the second side of the material or within the material;

where absence of the second signal on the second side of the material or within the material indicates that the modified living bacteria have not passed through or around the material and where presence of the second signal on the second side of the material or within the material indicates that the modified living bacteria have passed through or around the material.

4. The method of claim 1, where the first signal is light emission in the visible spectrum.

5. The method of claim 3, where the second signal is light emission in the visible spectrum.

6. (Amended) The method of claim 1, where the modified living bacteria are modified to incorporate a functional green fluorescent protein.

7. (Amended) The method of claim 1, where the modified living bacteria are modified to incorporate a functional luciferase.

8. (Amended) The method of claim 1, where the modified living bacteria are modified to incorporate both a functional green fluorescent protein and a functional luciferase.

9. (Amended) The method of claim 1, where placing the modified living bacteria on a first side of the material being evaluated comprises placing the modified living bacteria in the center of a hollowed out, extracted natural tooth where the root end of the tooth is sealed with the material, and then placing the root end of the tooth in a test medium; and

where [determining] detecting whether the first signal is present on a second side of the material or within the material comprises detecting the first signal in the test medium or within the material.

10. (Amended) The method of claim 3, where placing the modified living bacteria on a first side of the material being evaluated comprises placing the modified living bacteria in the center of a hollowed out, extracted natural tooth where the root end of the tooth is sealed with the material, and then placing the root end of the tooth in a test medium; and

where [determining] detecting whether the first signal is present on a second side of the material or within the material comprises detecting the first signal in the test medium or within the material.

11. (Amended) The method of claim 1, where the modified living bacteria provided are additionally modified to be grown selectively.

12. (Amended) The method of claim 11, where the modified living bacteria grow selectively due to antibiotic resistance.